

**Data Evaluation Report on the Acute Toxicity of Limonene to Aquatic Vascular Plants  
(*Lemna gibba* G3)**

PMRA Submission Number { ..... }

EPA MRID Number 49044001

**Data Requirement:** PMRA DATA CODE { ..... }  
EPA DP Barcode 409154  
OECD Data Point { ..... }  
EPA MRID 49044001  
EPA Guideline 850.4400

**Test material:** Avenger Weed Killer Concentrate      **Purity:** 70% a.i.  
Common name: Limonene  
Chemical name: IUPAC: Not reported  
                  CAS name: Not reported  
                  CAS No.: Not reported  
                  Synonyms:

**Primary Reviewer:** Kindra Bozicevich  
Environmental Scientist, CDM Smith

**Signature:** *Kindra Bozicevich*  
**Date:** 02/12/14

**Secondary Reviewer:** Teri S. Myers  
Environmental Scientist, CDM Smith

**Signature:** *Teri S. Myers*  
**Date:** 3/05/14

**Primary Reviewer:** Katherine Stebbins, EPA/OPP/ERB3

**Date:** 02/22/18 *Katherine Stebbins*

**EPA PC Code** 079701

**Date Evaluation Completed:** 02/22/18

**CITATION:** Mikulas, J. 2013. Avenger Weed Killer Concentrate Aquatic Plant Toxicity Tier II Test using *Lemna gibba*. Study performed by Stillmeadow, Inc., Sugar Land, TX. Study ID 16461-12. Study sponsored by Cutting Edge Formulation, Buford, GA and submitted by MacIntosh & Associates, Inc. Study initiated 02 October 2012 and completed 11 January 2013.

**DISCLAIMER:** This document provides guidance for EPA and PMRA reviewers on how to complete a data evaluation record after reviewing a scientific study concerning the acute toxicity of a pesticide to aquatic vascular plants. It is not intended to prescribe conditions to any external party for conducting this study nor to establish absolute criteria regarding the assessment of whether the study is scientifically sound and whether the study satisfies any applicable data requirements. Reviewers are expected to review and to determine for each study, on a case-by-case basis, whether it is scientifically sound and provides sufficient information to satisfy applicable data requirements. Studies that fail to meet any of the conditions may be accepted, if appropriate; similarly, studies that meet all of the conditions may be rejected, if appropriate. In sum, the reviewer is to take into account the totality of factors related to the test methodology and results in determining the acceptability of the study.

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## EXECUTIVE SUMMARY:

In a 7- day acute toxicity study, the freshwater floating aquatic vascular plants (duckweed, *Lemna gibba* G3) were exposed to Avenger Weed Killer Concentrate (70% ai d-limonene) at nominal concentrations of 0 (negative control) 3.125, 6.25, 12.5, 25, and 50 mg ai/L under static renewal conditions. Mean measured concentrations were not reported as the test concentrations were not analytically-determined. The NOAEC and EC50/IC50 values based on frond number yield were 3.125 and 29.65 mg ai/L, respectively, in terms of nominal concentrations. The NOAEC and EC50/IC50 values based on frond number growth rate were 3.125 and 69.51 mg ai/L, respectively, in terms of nominal concentrations. The NOAEC and EC50/IC50 values based on final biomass were 12.5 and 45.2 mg ai/L, respectively, in terms of nominal concentrations. The % growth inhibition of frond number in the treated culture as compared to the control ranged was 15.1 to 60.7%.

After 7 days, the fronds in the 25 mg/L test group appeared smaller and fronds in the 50 mg/L test group appeared small and had short roots. There was an increase in pH during the study.

The test concentrations were not analytically determined in this study and, as a result, all toxicity values are based upon nominal concentrations and the stability is not verified. Due to the volatility of the test compound, there is uncertainty with use of the nominal concentrations. Additionally, there is a lack of information describing the materials and methods used. Submission of data to support the stability of the formulation under test conditions and further description of any other methods used that could have potentially reduced the volatilization of a.i. over the test (e.g., closed flask), could possibly be used to upgrade the study classification. This study is classified as **SUPPLEMENTAL** and provides data that are useful for risk estimation and characterization and may be upgraded with further information to reduce the uncertainty with the exposure.

## Results Synopsis

Test Organism: Duckweed, *Lemna gibba* G3

Test Type (Flow-through, Static, Static Renewal): Static renewal

### *Frond number yield*

EC<sub>05</sub>: 1.213 mg ai/L 95% C.I.: N/A to 4.678 mg ai/L

EC<sub>50</sub>: 29.65 mg ai/L 95% C.I.: 19.84 to 44.32 mg ai/L

NOAEC: 3.125 mg ai/L

Probit Slope: N/A

### *Frond number growth rate*

EC<sub>05</sub>: 16.78 mg ai/L 95% C.I.: N/A to 24.83 mg ai/L

EC<sub>50</sub>: 69.51 mg ai/L 95% C.I.: 50.07 to 96.52 mg ai/L

NOAEC: 3.125 mg ai/L

Probit Slope: N/A

### *Final biomass*

EC<sub>05</sub>: 7.941 mg ai/L 95% C.I.: N/A to 12.72 mg ai/L

EC<sub>50</sub>: 45.2 mg ai/L 95% C.I.: 36.91 to 55.35 mg ai/L

NOAEC: 12.5 mg ai/L

Probit Slope: N/A

Endpoint(s) Effected: frond number yield, frond number growth rate, final biomass

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## I. MATERIALS AND METHODS

**GUIDELINE FOLLOWED:** The study was designed to comply with the procedures of the U.S. EPA OCSPP 850.4400 guideline. The following deviations from the U.S. EPA OCSPP 850.4400 (2012) guideline are noted:

1. The stability of the test material under test conditions was not assessed. As a result, mean measured test concentrations were not determined.
2. The original source of the test organism was not reported. It was noted that the stock cultures were maintained in-house, but the amount of time that the specific culture used in the test had been cultured in-house was not reported.
3. The health of the inoculum culture was not reported. Plants should be from a healthy stock culture.
4. The type of material used for the test containers and the fill volume was not reported.
5. The source/type, pH, total organic carbon (TOC), particulate matter, and chlorine concentrations of the dilution water were not reported.
6. The chelator used and the carbon source for the 20X AAP medium was not reported.
7. The method used to prepare the test solutions for the definitive test was not specified. The test material should be added to nutrient medium by either direct addition or by addition of a stock solution.
8. Only 3 replicates were used per control and treatment group. A minimum of 4 replicates per level is recommended.
9. The number of plants per replicate was not reported. It was only reported that 12-15 fronds were used per replicate. The same number of plants should be used in each replicate.
10. The test substance concentration was not reported at the start and end of each renewal period.
11. The test exposure concentrations were not analytically verified.

These deviations affect the validity of the study.

**COMPLIANCE:**

Signed and dated GLP, Quality Assurance and No Data Confidentiality statements were provided. The study was performed in accordance with the GLP standards of U.S. EPA (40 CFR Part 160) with the following exceptions: Section 160.31(d) and 160.105(a)(b)(e) Characterization and stability information was not provided to the testing facility. Section 160.113(a) Mixture analysis was not performed.

## **A. MATERIALS:**

**1. Test material** Avenger Weed Killer Concentrate

**Description:** Slight yellow liquid

**Lot No./Batch No. :** Lot# B186

**Purity:** 70% a.i.

**Stability of compound under test conditions:** Not assessed.

*(OECD recommends water solubility, stability in water and light, pKa, Pow, and vapor pressure of test compound)*

**Storage conditions of test chemicals:** Room temperature.

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## Physicochemical properties of Limonene.

Parameter	Values	Comments
Water solubility at 20EC	Not reported	
Vapor pressure	Not reported	
UV absorption	Not reported	
pKa	Not reported	
Kow	Not reported	

## 2. Test organism:

**Name:** Duckweed, *Lemna gibba* EPA requires a vascular species: *Lemna gibba*.

**Strain, if provided:** G3

**Source:** In-house cultures (original source not reported)

**Age of inoculum:** Mass culture initiated <2 weeks prior to dosing

**Method of cultivation:** Cultured and tested in 20X AAP medium under continuous light (4200 to 6700 lux) at 23-25°C.

## B. STUDY DESIGN:

### 1. Experimental Conditions

a. Range-finding study: A preliminary range-finding test was performed with test concentrations of 1, 10, 100, 500, and 1000 mg/L. The duckweed was introduced to each concentration at an initial frond count of 12-16, and at each renewal and test termination, the frond density was measured. Based on the results of the preliminary test, the definitive study was performed at test concentrations of 3.125, 6.25, 12.5, 25, and 50 mg/L.

b. Definitive Study

**Table 1: Experimental Parameters**

Parameter	Details	Remarks
		Criteria
Acclimation period:	Continuously cultured in-house	
Culturing media and conditions: (same as test or not)	Same as test	
Health: (any mortality observed)	None reported	
<u>Test system</u>		

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Parameter	Details	Remarks
		<i>Criteria</i>
Static/static renewal	Static renewal	<i>EPA expects the test concentrations to be renewed every 3 to 4 days (one renewal for the 7 day test, 3-4 renewals for the 14 day test).</i>
Renewal rate for static renewal	Renewed on days 3 and 5	
Incubation facility	Temperature-controlled cabinet held within an environmental chamber	
Duration of the test	7 days	<i>EPA requires a duration of 14 days. Seven day studies will be accepted for review by the Agency.</i>
<u>Test vessel</u> Material: ( <i>glass/stainless steel</i> ) Size: Fill volume:	Not reported 500 mL Not reported	Erlenmeyer flasks
<u>Details of 20X AAP medium</u> pH in new solutions: pH in old solutions: Chelator used: Carbon source:	7.6 to 7.9 8.2 to 8.5 Not reported Not reported	<i>EPA recommends the following culture media: Modified Hoagland's E+ or 20X-AAP. Chelating agents (e.g. EDTA) are recommended in the nutrient medium for optimum cell growth. Lower concentrations of chelating agents (down to one-third of the normal concentration recommended for AAP medium) may be used in the nutrient medium used for test solution preparation if it is suspected that the chelator will interact with the test material. ASTM reference, E1415-91 and D 3978-80 (reapproved 1987).</i>
If non-standard nutrient medium was used, detailed composition provided (Yes/No)	A standard medium was used. A detailed composition was not provided.	
<u>Dilution water</u>		

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Parameter	Details	Remarks
		Criteria
source/type: pH: water pretreatment (if any): Total Organic Carbon: particulate matter: metals: pesticides: chlorine:	Not reported Not reported Not reported Not reported Not reported None expected at concentrations considered to be toxic None expected at concentrations considered to be toxic Not reported	<i>EPA recommends a pH of ~5.0. A solution pH of 7.5 is acceptable if type 20X-AAP nutrient media is used.</i>
Indicate how the test material is added to the medium (added directly or used stock solution)	Not specified. In the preliminary study, the test material was added directly to the medium.	
Aeration or agitation	None	
<u>Sediment used (for rooted aquatic vascular plants)</u> Origin: Textural classification (% sand, silt, and clay): Organic carbon (%): Geographic location:	N/A	
<u>Number of replicates</u> Negative control: Solvent control: Treatments:	3 N/A 3	
Number of plants/replicate	Not reported	<i>EPA requires 5 plants.</i>
Number of fronds/plant	Not reported (initial frond count of 12-15)	<i>EPA requires 3 fronds per plant.</i>
<u>Test concentrations</u> Nominal:	0 (negative control), 3.125, 6.25, 12.5, 25, and 50 mg ai/L	TWA concentrations could not be calculated by the reviewer (see Reviewer's Comments)

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Parameter	Details	Remarks
		<i>Criteria</i>
Mean measured:	Not reported.	<i>EPA requires at least 5 test concentrations with a dose range of 2X or 3X progression.</i>
Solvent (type, percentage, if used)	N/A	
Method and interval of analytical verification	Not reported	
<u>Test conditions</u> Temperature: Photoperiod: Light intensity and quality:	23 to 25°C Continuous 4230 to 6830 lux fluorescent light	
<u>Reference chemical (if used)</u> name: concentrations:	N/A	
Other parameters, if any	N/A	

**2. Observations:**

**Table 2: Observation parameters**

Parameters	Details	Remarks/Criteria
Parameters measured (e.g.,: number of fronds, plant dry weight or other toxicity symptoms)	Frond number	
Measurement technique for frond number and other end points	Not reported	
Observation intervals	Fronds were counted on days 0, 3, 5, and 7.	
Other observations, if any	None	
Indicate whether there was an exponential growth in the control	Yes, the doubling time in the negative control was 2.4 days.	The reviewer calculated the doubling time of the negative control as $\ln(2)/0.291$ . 0.291 is the 0-7 day frond number growth rate of the negative control.
Were raw data included?	Yes	

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**II. RESULTS and DISCUSSION:**

**A. INHIBITORY EFFECTS:**

After 7 days, the mean frond number of the negative control was 106, yielding inhibitions relative to the negative control of 15.1, 31.1, 26.7, 34.9, and 60.7% for nominal concentrations of 3.125, 6.25, 12.5, 25, and 50 mg ai/L. The NOAEC and EC50 values reported by the study author based on final frond number were 3.125 and 39.634 mg ai/L, respectively, in terms of nominal concentrations.

The mean 0-7 day frond number growth rate of the negative control was 0.291/day, yielding inhibitions relative to the negative control of 5.3, 11.6, 11.2, 15.0, and 39.6% for nominal concentrations of 3.125, 6.25, 12.5, 25, and 50 mg ai/L. The NOAEC and EC50 values reported by the study author based on frond number growth rate were 12.5 and >50 mg ai/L, respectively, in terms of nominal concentrations.

The mean 0-7 day frond number yield of the negative control was 92 fronds, yielding inhibitions relative to the negative control of 16.6, 33.9, 29.6, 38.3, and 67.9% for nominal concentrations of 3.125, 6.25, 12.5, 25, and 50 mg ai/L. The study author did not assess frond number yield.

After 7 days, the mean final dry weight biomass of the negative control was 0.1703 g, yielding inhibitions relative to the negative control of -2.7, -8.6, 4.4, 30.7, and 49.9% for nominal concentration of 3.125, 6.25, 12.5, 25, and 50 mg ai/L. The NOAEC and EC50 values reported by the study author based on mean biomass were 12.5 and 47.695 mg ai/L, respectively, in terms of nominal concentrations.

The dry weight biomass yield and the mean 0-7 day dry weight biomass growth rate could not be calculated. The reviewer did not report the initial frond weight.

After 7 days, the fronds in the 25 mg/L test group appeared smaller and fronds in the 50 mg/L test group appeared small and had short roots. There was an increase in pH during the study.

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**Table 3: Effect of Avenger Weed Killer Concentrate on frond number of duckweed, *Lemna gibba***

Treatment Nominal concentrations mg ai/L	Initial frond number/test solution	frond number at			
		3 days	5 days	7 days	
				frond number	% inhibition <sup>a</sup>
Negative control	12-15	30	75	106	N/A
3.125	12-14	28	69	90	15.1
6.25	12	23	57	73	31.1
12.5	12-14	28	63	78	26.7
25	12	25	58	69	34.9
50	12	23	33	42	60.7
Reference chemical (if used)	N/A				

<sup>a</sup> Calculated by the reviewer relative to the negative control.

**Table 4: Effect of Avenger Weed Killer Concentrate on frond number of duckweed, *Lemna gibba***

Treatment Nominal concentrations mg ai/L	Initial frond number/test solution (or other endpoint)	Frond number growth rate (day <sup>-1</sup> ) <sup>a,b</sup>		Frond number yield <sup>c</sup>	
		0-7 days	% inhibition <sup>d</sup>	0-7 days	% inhibition <sup>d</sup>
Negative control	12-15	0.291	N/A	92	N/A
3.125	12-14	0.276	5.3	77	16.6
6.25	12	0.258	11.6	61	33.9
12.5	12-14	0.259	11.2	65	29.6
25	12	0.248	15.0	57	38.3
50	12	0.176	39.6	30	67.9

<sup>a</sup>Calculated by the reviewer using a logarithmic growth equation.

<sup>b</sup> Values reported here are those calculated by the reviewer, as the reviewer's calculated frond number growth rate values differed slightly from those of the study author. The mean frond number growth rate values reported by the study author were 0.0283, 0.0268, 0.0251, 0.0252, 0.0241, and 0.0171 for the negative control and nominal concentrations of 3.125, 6.25, 12.5, 25, and 50 mg/L, respectively. The study author did not specify units for these values.

<sup>c</sup> Calculated by the reviewer as final minus initial frond number.

<sup>d</sup> Calculated by the reviewer relative to the negative control.

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**Table 5: Effect of Avenger Weed Killer Concentrate on biomass of duckweed, *Lemna gibba***

Treatment Nominal concentrations mg ai/L	Initial frond weight/test solution (mg)	Final biomass (g)	
		Day 7	% inhibition <sup>a</sup>
Negative control	NR	0.1703	N/A
3.125	NR	0.1749	-2.7
6.25	NR	0.1850	-8.6
12.5	NR	0.1628	4.4
25	NR	0.1180	30.7
50	NR	0.0854	49.9

<sup>a</sup> Calculated by the reviewer relative to the negative control.

**Table 6: Statistical endpoint values.\* (calculated by the study author based on nominal concentrations)**

Statistical Endpoint	Frond number (final)	Frond number yield	Frond number growth rate	Biomass (final)
NOAEC (mg ai/L)	3.125	Not calculated	12.5	12.5
LOAEC (mg ai/L)	6.25	Not calculated	25.0	25.0
IC <sub>05</sub> or EC <sub>05</sub> (mg ai/L) (95% C.I.)	1.035 (0.000- 7.345)	Not calculated	2.927 (0.000- 24.260)	10.199 (0.000- 19.342)
IC <sub>50</sub> or EC <sub>50</sub> (mg ai/L) (95% C.I.)	39.634 (N/A)	Not calculated	>50	47.695
Reference chemical NOAEC IC <sub>50</sub> /EC <sub>50</sub>	N/A			

\* Do not use this table, if the study was deemed unacceptable.

N/A. Not applicable.

**B. REPORTED STATISTICS:**

The study author statistically analyzed all endpoints using ToxCalc Version 5.0™ statistical software. The EC values were determined using one of the following methods: Probit, Trimmed Spearman-Karber, and Linear Interpolation. All analyses were based on nominal concentrations.

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## C. VERIFICATION OF STATISTICAL RESULTS:

**Statistical Method:** The reviewer assessed the endpoints for frond number yield, frond number growth rate, and final biomass using CETIS version 1.8.7.12 statistical software with backend database settings implemented by EFED on 31 May 2013. The data were all confirmed to be normally distributed and have homogeneous variances using Shapiro-Wilk's and Bartlett's tests, respectively, and were therefore analyzed using ANOVA followed by Dunnett's tests. The ECx values were determined using Bruce-Versteeg regression. All analyses were based on nominal concentrations.

### *Frond number yield*

EC <sub>05</sub> :	1.213 mg ai/L	95% C.I.: N/A to 4.678 mg ai/L
EC <sub>50</sub> :	29.65 mg ai/L	95% C.I.: 19.84 to 44.32 mg ai/L
NOAEC:	3.125 mg ai/L	

Probit Slope: N/A

### *Frond number growth rate*

EC <sub>05</sub> :	16.78 mg ai/L	95% C.I.: N/A to 24.83 mg ai/L
EC <sub>50</sub> :	69.51 mg ai/L	95% C.I.: 50.07 to 96.52 mg ai/L
NOAEC:	3.125 mg ai/L	

Probit Slope: N/A

### *Final biomass*

EC <sub>05</sub> :	7.941 mg ai/L	95% C.I.: N/A to 12.72 mg ai/L
EC <sub>50</sub> :	45.2 mg ai/L	95% C.I.: 36.91 to 55.35 mg ai/L
NOAEC:	12.5 mg ai/L	

Probit Slope: N/A

## D. STUDY DEFICIENCIES:

The test concentrations were not analytically-determined in this study and, as a result, all toxicity values are based upon nominal concentrations. This deficiency along with the use of an End use product (70% a.i.), rather than TGAI led to a classification of SUPPLEMENTAL. There were also several other guideline deviations and reporting deficiencies noted in the Materials and Methods section of this DER.

## E. REVIEWER'S COMMENTS:

The reviewer's results were in general agreement with those of the study author, except that the reviewer's analysis (given the suggestively-monotonic decreasing trend) detected a lower NOAEC than the study author's for frond number growth rate. The reviewer's results are presented in the Executive Summary and Conclusions sections of this report.

The test solutions were renewed on Days 3 and 5; however, the study author did not report measurement of the exposure concentrations.

The laboratory portion of the definitive test was conducted from 05 to 21 November 2012.

## F. CONCLUSIONS:

This study is classified as SUPPLEMENTAL. After 7 days, the most sensitive endpoint was frond number yield with EC<sub>50</sub> and NOAEC values of 29.65 and 3.125 mg ai/L, respectively, based on nominal concentrations.

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*Frond number yield*

EC<sub>05</sub>: 1.213 mg ai/L      95% C.I.: N/A to 4.678 mg ai/L  
EC<sub>50</sub>: 29.65 mg ai/L      95% C.I.: 19.84 to 44.32 mg ai/L  
NOAEC: 3.125 mg ai/L

Probit Slope: N/A

*Frond number growth rate*

EC<sub>05</sub>: 16.78 mg ai/L      95% C.I.: N/A to 24.83 mg ai/L  
EC<sub>50</sub>: 69.51 mg ai/L      95% C.I.: 50.07 to 96.52 mg ai/L  
NOAEC: 3.125 mg ai/L

Probit Slope: N/A

*Final biomass*

EC<sub>05</sub>: 7.941 mg ai/L      95% C.I.: N/A to 12.72 mg ai/L  
EC<sub>50</sub>: 45.2 mg ai/L      95% C.I.: 36.91 to 55.35 mg ai/L  
NOAEC: 12.5 mg ai/L

Probit Slope: N/A

Endpoint(s) Effected: frond number yield, frond number growth rate, final biomass

**III. REFERENCES:** None reported.

**CETIS Summary Report**

**Report Date:** 11 Feb-14 12:11 (p 1 of 2)  
**Test Code:** 079701 49044001 | 01-3500-9253

**OCSPP 850.4400 Aquatic Vascular Plant****Stillmeadow, Inc.**

<b>Batch ID:</b> 03-7100-1831	<b>Test Type:</b> Lemna Growth (7-d)	<b>Analyst:</b>
<b>Start Date:</b> 05 Nov-12	<b>Protocol:</b> OCSPP 850.4400 Aquatic Plant (Lemna)	<b>Diluent:</b> Lemna Medium, 20X-AAP
<b>Ending Date:</b> 21 Nov-12	<b>Species:</b> Lemna Gibba	<b>Brine:</b>
<b>Duration:</b> 16d 0h	<b>Source:</b> Lab In-House Culture	<b>Age:</b>
<b>Sample ID:</b> 21-0260-0787	<b>Code:</b> 49044001	<b>Client:</b> CDM Smith
<b>Sample Date:</b> 05 Nov-12	<b>Material:</b> Limonene	<b>Project:</b> Unknown
<b>Receive Date:</b>	<b>Source:</b> MacIntosh & Associates (MacIntosh)	
<b>Sample Age:</b> NA	<b>Station:</b>	

**Batch Note:** 079701 49044001 static renewal**Sample Note:** 079701 49044001**Comparison Summary**

<b>Analysis ID</b>	<b>Endpoint</b>	<b>NOEL</b>	<b>LOEL</b>	<b>TOEL</b>	<b>PMSD</b>	<b>TU</b>	<b>Method</b>
11-3391-6644	Frond Number	3.125	6.25	4.419	26.4%		Dunnett Multiple Comparison Test
04-6712-2144	Frond Number	3.125	6.25	4.419	20.3%		Williams Multiple Comparison Test
02-5927-7286	Frond Number Growth Rate	12.5	25	17.68	14.8%		Dunnett Multiple Comparison Test
10-3489-0666	Frond Number Growth Rate	3.125	6.25	4.419	11.4%		Williams Multiple Comparison Test
19-4868-2227	Frond Weight	12.5	25	17.68	21.9%		Dunnett Multiple Comparison Test
05-7312-0626	Frond Weight	12.5	25	17.68	16.9%		Williams Multiple Comparison Test

**Point Estimate Summary**

<b>Analysis ID</b>	<b>Endpoint</b>	<b>Level</b>	<b>mg ai/L</b>	<b>95% LCL</b>	<b>95% UCL</b>	<b>TU</b>	<b>Method</b>
04-1108-0388	Frond Number	IC5	1.213	N/A	4.678		Nonlinear Regression
		IC10	2.457	N/A	6.174		
		IC25	7.994	3.982	13.97		
		IC50	29.65	19.84	44.32		
10-1825-1279	Frond Number Growth Rat	IC5	16.78	N/A	24.83		Nonlinear Regression
		IC10	22.97	11.48	31.66		
		IC25	38.81	31.81	46.09		
		IC50	69.51	50.07	96.52		
10-8444-5970	Frond Weight	IC5	7.941	N/A	12.72		Nonlinear Regression
		IC10	11.66	5.347	17.05		
		IC25	22.15	16.32	28.58		
		IC50	45.2	36.91	55.35		

**CETIS Summary Report**

 Report Date: 11 Feb-14 12:11 (p 2 of 2)  
 Test Code: 079701 49044001 | 01-3500-9253

**OCSPP 850.4400 Aquatic Vascular Plant**
**Stillmeadow, Inc.**
**Frond Number Summary**

C-mg ai/L	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	Negative Control	3	92.33	43.63	141	74	113	11.32	19.6	21.23%	0.0%
3.125		3	77	51.06	102.9	65	84	6.028	10.44	13.56%	16.61%
6.25		3	61	48.58	73.42	56	66	2.887	5	8.2%	33.94%
12.5		3	65	43.78	86.22	57	74	4.933	8.544	13.14%	29.6%
25		3	57	22.22	91.78	41	67	8.083	14	24.56%	38.27%
50		3	29.67	9.435	49.9	24	39	4.702	8.145	27.45%	67.87%

**Frond Number Growth Rate Summary**

C-mg ai/L	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	Negative Control	3	0.2913	0.2403	0.3424	0.278	0.315	0.01186	0.02055	7.05%	0.0%
3.125		3	0.276	0.2532	0.2988	0.266	0.284	0.005292	0.009165	3.32%	5.26%
6.25		3	0.2577	0.2341	0.2813	0.248	0.267	0.005487	0.009504	3.69%	11.56%
12.5		3	0.2587	0.2102	0.3071	0.245	0.281	0.01126	0.0195	7.54%	11.21%
25		3	0.2477	0.1704	0.3249	0.212	0.269	0.01795	0.03109	12.55%	14.99%
50		3	0.1763	0.1096	0.2431	0.157	0.207	0.01551	0.02686	15.23%	39.47%

**Frond Weight Summary**

C-mg ai/L	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	Negative Control	3	0.1703	0.08341	0.2573	0.1431	0.2098	0.0202	0.03499	20.54%	0.0%
3.125		3	0.1749	0.1604	0.1895	0.1706	0.1816	0.003383	0.005859	3.35%	-2.7%
6.25		3	0.185	0.1714	0.1986	0.1812	0.1913	0.003172	0.005495	2.97%	-8.61%
12.5		3	0.1628	0.1169	0.2086	0.1481	0.1835	0.01066	0.01846	11.34%	4.44%
25		3	0.118	0.1029	0.1332	0.1133	0.1249	0.003514	0.006087	5.16%	30.7%
50		3	0.08537	0.04013	0.1306	0.0675	0.1039	0.01051	0.01821	21.33%	49.88%

**Frond Number Detail**

C-mg ai/L	Control Type	Rep 1	Rep 2	Rep 3
0	Negative Control	74	90	113
3.125		82	84	65
6.25		66	61	56
12.5		57	64	74
25		67	41	63
50		26	39	24

**Frond Number Growth Rate Detail**

C-mg ai/L	Control Type	Rep 1	Rep 2	Rep 3
0	Negative Control	0.281	0.278	0.315
3.125		0.284	0.278	0.266
6.25		0.267	0.258	0.248
12.5		0.25	0.245	0.281
25		0.269	0.212	0.262
50		0.165	0.207	0.157

**Frond Weight Detail**

C-mg ai/L	Control Type	Rep 1	Rep 2	Rep 3
0	Negative Control	0.2098	0.1581	0.1431
3.125		0.1726	0.1706	0.1816
6.25		0.1913	0.1825	0.1812
12.5		0.1835	0.1567	0.1481
25		0.1159	0.1249	0.1133
50		0.0847	0.0675	0.1039

# CETIS Analytical Report

Report Date: 11 Feb-14 12:00 (p 1 of 12)  
 Test Code: 079701 49044001 | 01-3500-9253

OCSPP 850.4400 Aquatic Vascular Plant								Stillmeadow, Inc.					
Analysis ID:		Endpoint: Frond Number Growth Rate				CETIS Version:		CETISv1.8.7					
Analyzed:		Analysis: Parametric-Control vs Treatments				Official Results:		Yes					
Batch ID:	03-7100-1831	Test Type: Lemna Growth (7-d)				Analyst:							
Start Date:	05 Nov-12	Protocol: OCSPP 850.4400 Aquatic Plant (Lemna)				Diluent: Lemna Medium, 20X-AAP							
Ending Date:	21 Nov-12	Species: Lemna Gibba				Brine:							
Duration:	16d 0h	Source: Lab In-House Culture				Age:							
Data Transform	Zeta	Alt Hyp	Trials	Seed	PMSD	NOEL	LOEL	TOEL	TU				
Untransformed	NA	C > T	NA	NA	14.8%	12.5	25	17.68					
<b>Dunnett Multiple Comparison Test</b>													
Control	vs	C-mg ai/L	Test Stat	Critical	MSD	DF	P-Value	P-Type	Decision( $\alpha:5\%$ )				
Negative Control	3.125	0.8911	2.502	0.043	4	0.4738	CDF	Non-Significant Effect					
	6.25	1.957	2.502	0.043	4	0.1218	CDF	Non-Significant Effect					
	12.5	1.898	2.502	0.043	4	0.1332	CDF	Non-Significant Effect					
	25*	2.538	2.502	0.043	4	0.0471	CDF	Significant Effect					
	50*	6.683	2.502	0.043	4	<0.0001	CDF	Significant Effect					
<b>ANOVA Table</b>													
Source	Sum Squares		Mean Square		DF	F Stat		P-Value	Decision( $\alpha:5\%$ )				
Between	0.02382228		0.004764455		5	10.73		0.0004	Significant Effect				
Error	0.005329333		0.0004441111		12								
Total	0.02915161				17								
<b>Distributional Tests</b>													
Attribute	Test		Test Stat	Critical	P-Value	Decision( $\alpha:1\%$ )							
Variances	Bartlett Equality of Variance		3.685	15.09	0.5956	Equal Variances							
Variances	Mod Levene Equality of Variance		0.3208	8.746	0.8836	Equal Variances							
Variances	Levene Equality of Variance		2.53	5.064	0.0872	Equal Variances							
Distribution	Shapiro-Wilk W Normality		0.9532	0.8546	0.4768	Normal Distribution							
Distribution	Kolmogorov-Smirnov D		0.1878	0.2344	0.0933	Normal Distribution							
Distribution	D'Agostino Skewness		0.1083	2.576	0.9138	Normal Distribution							
Distribution	Anderson-Darling A2 Normality		0.4955	3.878	0.2181	Normal Distribution							
<b>Frond Number Growth Rate Summary</b>													
C-mg ai/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect		
0	Negative Control	3	0.2913	0.2403	0.3424	0.281	0.278	0.315	0.01186	7.05%	0.0%		
3.125		3	0.276	0.2532	0.2988	0.278	0.266	0.284	0.005292	3.32%	5.26%		
6.25		3	0.2577	0.2341	0.2813	0.258	0.248	0.267	0.005487	3.69%	11.56%		
12.5		3	0.2587	0.2102	0.3071	0.25	0.245	0.281	0.01126	7.54%	11.21%		
25		3	0.2477	0.1704	0.3249	0.262	0.212	0.269	0.01795	12.55%	14.99%		
50		3	0.1763	0.1096	0.2431	0.165	0.157	0.207	0.01551	15.23%	39.47%		

# CETIS Analytical Report

Report Date: 11 Feb-14 12:00 (p 2 of 12)  
Test Code: 079701 49044001 | 01-3500-9253

## OCSPP 850.4400 Aquatic Vascular Plant

Stillmeadow, Inc.

Analysis ID: 02-5927-7286

Endpoint: Frond Number Growth Rate

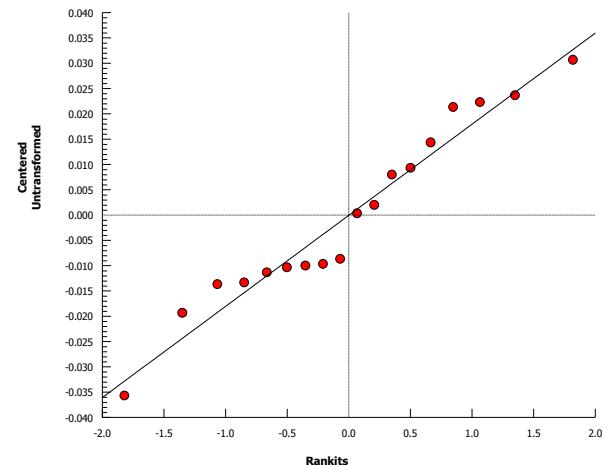
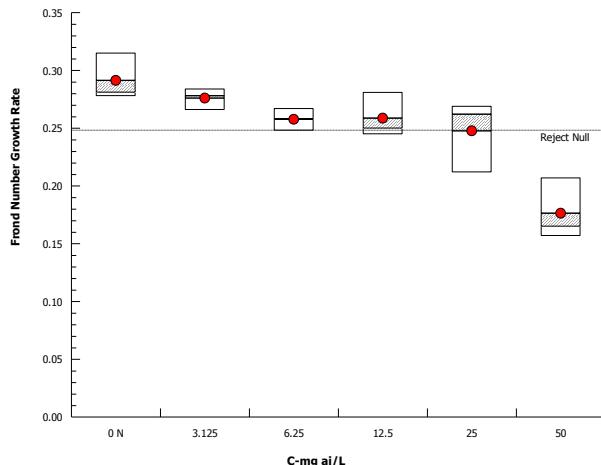
CETIS Version: CETISv1.8.7

Analyzed: 11 Feb-14 11:57

Analysis: Parametric-Control vs Treatments

Official Results: Yes

### Graphics



# CETIS Analytical Report

Report Date: 11 Feb-14 12:00 (p 3 of 12)  
 Test Code: 079701 49044001 | 01-3500-9253

## OCSPP 850.4400 Aquatic Vascular Plant Stillmeadow, Inc.

Analysis ID:	10-3489-0666	Endpoint:	Frond Number Growth Rate	CETIS Version:	CETISv1.8.7
Analyzed:	11 Feb-14 11:57	Analysis:	Parametric-Control vs Ord.Treatments	Official Results:	Yes
Batch ID:	03-7100-1831	Test Type:	Lemna Growth (7-d)	Analyst:	
Start Date:	05 Nov-12	Protocol:	OCSPP 850.4400 Aquatic Plant (Lemna)	Diluent:	Lemna Medium, 20X-AAP
Ending Date:	21 Nov-12	Species:	Lemna Gibba	Brine:	
Duration:	16d 0h	Source:	Lab In-House Culture	Age:	
Data Transform	Zeta	Alt Hyp	Trials	Seed	PMSD NOEL LOEL TOEL TU
Untransformed	NA	C > T	NA	NA	11.4% 3.125 6.25 4.419

### Williams Multiple Comparison Test

Control	vs	C-mg ai/L	Test Stat	Critical	MSD	DF	P-Value	P-Type	Decision( $\alpha:5\%$ )
Negative Control		3.125	0.8911	1.782	0.031	4	>0.05	CDF	Non-Significant Effect
		6.25*	1.957	1.873	0.032	4	<0.05	CDF	Significant Effect
		12.5*	1.928	1.903	0.033	4	<0.05	CDF	Significant Effect
		25*	2.538	1.918	0.033	4	<0.05	CDF	Significant Effect
		50*	6.683	1.927	0.033	4	<0.05	CDF	Significant Effect

### ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision( $\alpha:5\%$ )
Between	0.02382228	0.004764455	5	10.73	0.0004	Significant Effect
Error	0.005329333	0.0004441111	12			
Total	0.02915161		17			

### Distributional Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision( $\alpha:1\%$ )
Variances	Bartlett Equality of Variance	3.685	15.09	0.5956	Equal Variances
Variances	Mod Levene Equality of Variance	0.3208	8.746	0.8836	Equal Variances
Variances	Levene Equality of Variance	2.53	5.064	0.0872	Equal Variances
Distribution	Shapiro-Wilk W Normality	0.9532	0.8546	0.4768	Normal Distribution
Distribution	Kolmogorov-Smirnov D	0.1878	0.2344	0.0933	Normal Distribution
Distribution	D'Agostino Skewness	0.1083	2.576	0.9138	Normal Distribution
Distribution	Anderson-Darling A2 Normality	0.4955	3.878	0.2181	Normal Distribution

### Frond Number Growth Rate Summary

C-mg ai/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Negative Control	3	0.2913	0.2403	0.3424	0.281	0.278	0.315	0.01186	7.05%	0.0%
3.125		3	0.276	0.2532	0.2988	0.278	0.266	0.284	0.005292	3.32%	5.26%
6.25		3	0.2577	0.2341	0.2813	0.258	0.248	0.267	0.005487	3.69%	11.56%
12.5		3	0.2587	0.2102	0.3071	0.25	0.245	0.281	0.01126	7.54%	11.21%
25		3	0.2477	0.1704	0.3249	0.262	0.212	0.269	0.01795	12.55%	14.99%
50		3	0.1763	0.1096	0.2431	0.165	0.157	0.207	0.01551	15.23%	39.47%

# CETIS Analytical Report

Report Date: 11 Feb-14 12:00 (p 4 of 12)  
Test Code: 079701 49044001 | 01-3500-9253

## OCSPP 850.4400 Aquatic Vascular Plant

Stillmeadow, Inc.

Analysis ID: 10-3489-0666

Endpoint: Frond Number Growth Rate

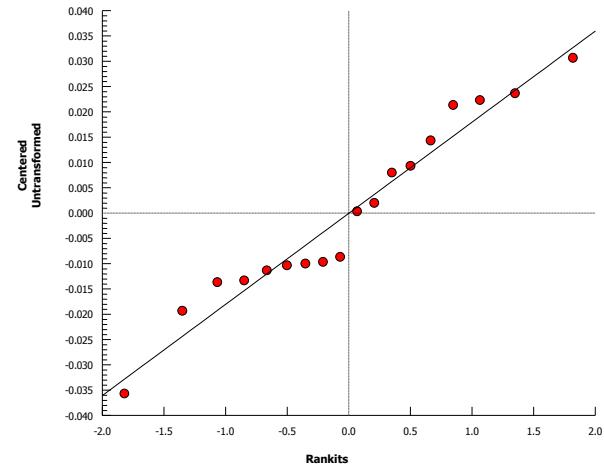
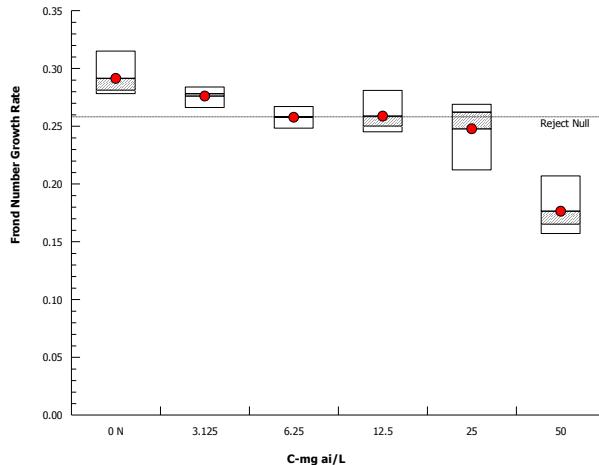
CETIS Version: CETISv1.8.7

Analyzed: 11 Feb-14 11:57

Analysis: Parametric-Control vs Ord.Treatments

Official Results: Yes

### Graphics



# CETIS Analytical Report

Report Date: 11 Feb-14 12:00 (p 5 of 12)  
 Test Code: 079701 49044001 | 01-3500-9253

OCSPP 850.4400 Aquatic Vascular Plant								Stillmeadow, Inc.					
Analysis ID:		Endpoint: Frond Number				CETIS Version:		CETISv1.8.7					
Analyzed:		Analysis: Parametric-Control vs Treatments				Official Results:		Yes					
Batch ID:	03-7100-1831	Test Type: Lemna Growth (7-d)				Analyst:							
Start Date:	05 Nov-12	Protocol: OCSPP 850.4400 Aquatic Plant (Lemna)				Diluent: Lemna Medium, 20X-AAP							
Ending Date:	21 Nov-12	Species: Lemna Gibba				Brine:							
Duration:	16d 0h	Source: Lab In-House Culture				Age:							
Data Transform	Zeta	Alt Hyp	Trials	Seed	PMSD	NOEL	LOEL	TOEL	TU				
Untransformed	NA	C > T	NA	NA	26.4%	3.125	6.25	4.419					
<b>Dunnett Multiple Comparison Test</b>													
Control	vs	C-mg ai/L	Test Stat	Critical	MSD	DF	P-Value	P-Type	Decision( $\alpha:5\%$ )				
Negative Control		3.125	1.574	2.502	24.37	4	0.2132	CDF	Non-Significant Effect				
		6.25*	3.217	2.502	24.37	4	0.0143	CDF	Significant Effect				
		12.5*	2.807	2.502	24.37	4	0.0296	CDF	Significant Effect				
		25*	3.628	2.502	24.37	4	0.0069	CDF	Significant Effect				
		50*	6.434	2.502	24.37	4	<0.0001	CDF	Significant Effect				
<b>ANOVA Table</b>													
Source	Sum Squares		Mean Square		DF	F Stat		P-Value	Decision( $\alpha:5\%$ )				
Between	6626.667		1325.333		5	9.315		0.0008	Significant Effect				
Error	1707.333		142.2778		12								
Total	8334				17								
<b>Distributional Tests</b>													
Attribute	Test		Test Stat	Critical	P-Value	Decision( $\alpha:1\%$ )							
Variances	Bartlett Equality of Variance		3.553	15.09	0.6154	Equal Variances							
Variances	Mod Levene Equality of Variance		0.867	8.746	0.5526	Equal Variances							
Variances	Levene Equality of Variance		1.441	5.064	0.2791	Equal Variances							
Distribution	Shapiro-Wilk W Normality		0.9767	0.8546	0.9095	Normal Distribution							
Distribution	Kolmogorov-Smirnov D		0.1355	0.2344	0.5354	Normal Distribution							
Distribution	D'Agostino Skewness		0.08981	2.576	0.9284	Normal Distribution							
Distribution	Anderson-Darling A2 Normality		0.244	3.878	0.7908	Normal Distribution							
<b>Frond Number Summary</b>													
C-mg ai/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect		
0	Negative Control	3	92.33	43.63	141	90	74	113	11.32	21.23%	0.0%		
3.125		3	77	51.06	102.9	82	65	84	6.028	13.56%	16.61%		
6.25		3	61	48.58	73.42	61	56	66	2.887	8.2%	33.94%		
12.5		3	65	43.78	86.22	64	57	74	4.933	13.14%	29.6%		
25		3	57	22.22	91.78	63	41	67	8.083	24.56%	38.27%		
50		3	29.67	9.435	49.9	26	24	39	4.702	27.45%	67.87%		

# CETIS Analytical Report

Report Date: 11 Feb-14 12:00 (p 6 of 12)  
Test Code: 079701 49044001 | 01-3500-9253

## OCSPP 850.4400 Aquatic Vascular Plant

Stillmeadow, Inc.

Analysis ID: 11-3391-6644

Endpoint: Frond Number

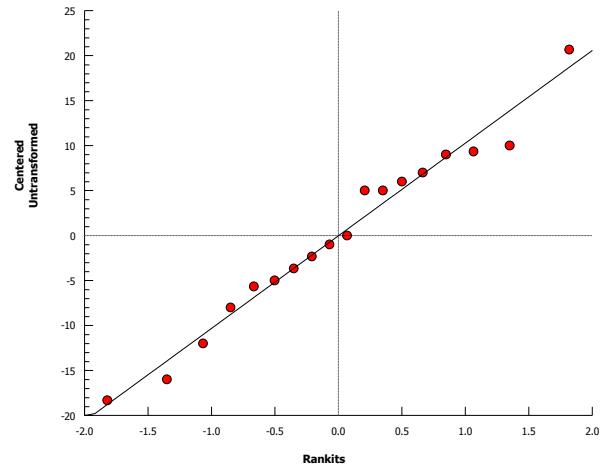
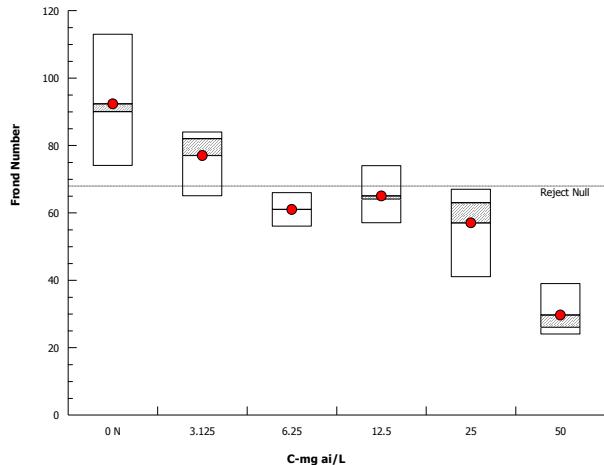
CETIS Version: CETISv1.8.7

Analyzed: 11 Feb-14 11:57

Analysis: Parametric-Control vs Treatments

Official Results: Yes

### Graphics



# CETIS Analytical Report

Report Date: 11 Feb-14 12:00 (p 7 of 12)  
 Test Code: 079701 49044001 | 01-3500-9253

OCSPP 850.4400 Aquatic Vascular Plant								Stillmeadow, Inc.							
Analysis ID:		04-6712-2144	Endpoint: Frond Number				CETIS Version:		CETISv1.8.7						
Analyzed:		11 Feb-14 11:57	Analysis: Parametric-Control vs Ord.Treatments				Official Results:		Yes						
Batch ID:	03-7100-1831	Test Type: Lemna Growth (7-d)				Analyst:									
Start Date:	05 Nov-12	Protocol: OCSPP 850.4400 Aquatic Plant (Lemna)				Diluent: Lemna Medium, 20X-AAP									
Ending Date:	21 Nov-12	Species: Lemna Gibba				Brine:									
Duration:	16d 0h	Source: Lab In-House Culture				Age:									
Data Transform	Zeta	Alt Hyp	Trials	Seed		PMSD	NOEL	LOEL	TOEL	TU					
Untransformed	NA	C > T	NA	NA		20.3%	3.125	6.25	4.419						
<b>Williams Multiple Comparison Test</b>															
Control	vs	C-mg ai/L	Test Stat	Critical	MSD	DF	P-Value	P-Type	Decision( $\alpha:5\%$ )						
Negative Control	3.125	1.574	1.782	17.36	4	>0.05	CDF	Non-Significant Effect							
	6.25*	3.217	1.873	18.24	4	<0.05	CDF	Significant Effect							
	12.5*	3.012	1.903	18.53	4	<0.05	CDF	Significant Effect							
	25*	3.628	1.918	18.68	4	<0.05	CDF	Significant Effect							
	50*	6.434	1.927	18.77	4	<0.05	CDF	Significant Effect							
<b>ANOVA Table</b>															
Source	Sum Squares		Mean Square		DF	F Stat	P-Value	Decision( $\alpha:5\%$ )							
Between	6626.667		1325.333		5	9.315	0.0008	Significant Effect							
Error	1707.333		142.2778		12										
Total	8334				17										
<b>Distributional Tests</b>															
Attribute	Test		Test Stat	Critical	P-Value	Decision( $\alpha:1\%$ )									
Variances	Bartlett Equality of Variance		3.553	15.09	0.6154	Equal Variances									
Variances	Mod Levene Equality of Variance		0.867	8.746	0.5526	Equal Variances									
Variances	Levene Equality of Variance		1.441	5.064	0.2791	Equal Variances									
Distribution	Shapiro-Wilk W Normality		0.9767	0.8546	0.9095	Normal Distribution									
Distribution	Kolmogorov-Smirnov D		0.1355	0.2344	0.5354	Normal Distribution									
Distribution	D'Agostino Skewness		0.08981	2.576	0.9284	Normal Distribution									
Distribution	Anderson-Darling A2 Normality		0.244	3.878	0.7908	Normal Distribution									
<b>Frond Number Summary</b>															
C-mg ai/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect				
0	Negative Control	3	92.33	43.63	141	90	74	113	11.32	21.23%	0.0%				
3.125		3	77	51.06	102.9	82	65	84	6.028	13.56%	16.61%				
6.25		3	61	48.58	73.42	61	56	66	2.887	8.2%	33.94%				
12.5		3	65	43.78	86.22	64	57	74	4.933	13.14%	29.6%				
25		3	57	22.22	91.78	63	41	67	8.083	24.56%	38.27%				
50		3	29.67	9.435	49.9	26	24	39	4.702	27.45%	67.87%				

# CETIS Analytical Report

Report Date: 11 Feb-14 12:00 (p 8 of 12)  
Test Code: 079701 49044001 | 01-3500-9253

## OCSPP 850.4400 Aquatic Vascular Plant

Stillmeadow, Inc.

Analysis ID: 04-6712-2144

Endpoint: Frond Number

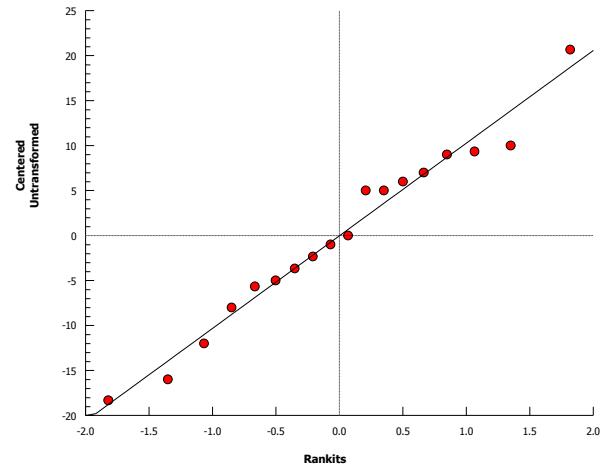
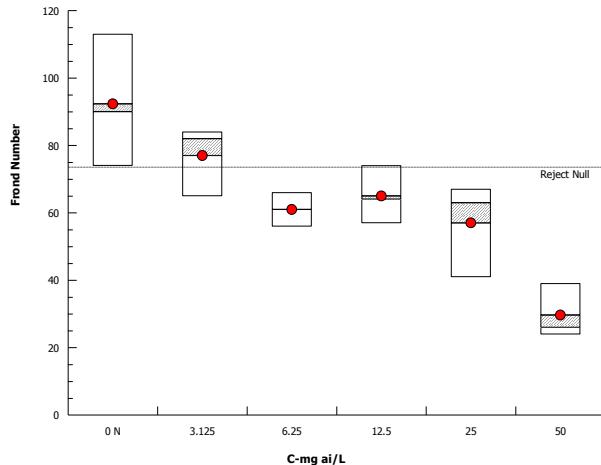
CETIS Version: CETISv1.8.7

Analyzed: 11 Feb-14 11:57

Analysis: Parametric-Control vs Ord.Treatments

Official Results: Yes

### Graphics



# CETIS Analytical Report

Report Date: 11 Feb-14 12:00 (p 9 of 12)  
 Test Code: 079701 49044001 | 01-3500-9253

OCSPP 850.4400 Aquatic Vascular Plant								Stillmeadow, Inc.				
Analysis ID:		19-4868-2227	Endpoint:			Frond Weight		CETIS Version:		CETISv1.8.7		
Analyzed:		11 Feb-14 11:57	Analysis:			Parametric-Control vs Treatments		Official Results:		Yes		
Batch ID:	03-7100-1831	Test Type:			Lemna Growth (7-d)			Analyst:				
Start Date:	05 Nov-12	Protocol:			OCSPP 850.4400 Aquatic Plant (Lemna)			Diluent:				
Ending Date:	21 Nov-12	Species:			Lemna Gibba			Brine:				
Duration:	16d 0h	Source:			Lab In-House Culture			Age:				
Data Transform	Zeta	Alt Hyp	Trials	Seed	PMSD	NOEL	LOEL	TOEL	TU			
Untransformed	NA	C > T	NA	NA	21.9%	12.5	25	17.68				
<b>Dunnett Multiple Comparison Test</b>												
Control	vs	C-mg ai/L	Test Stat	Critical	MSD	DF	P-Value	P-Type	Decision( $\alpha:5\%$ )			
Negative Control	3.125	-0.3087	2.502	0.037	4	0.9076	CDF	Non-Significant Effect				
	6.25	-0.9842	2.502	0.037	4	0.9808	CDF	Non-Significant Effect				
	12.5	0.5078	2.502	0.037	4	0.6454	CDF	Non-Significant Effect				
	25*	3.51	2.502	0.037	4	0.0085	CDF	Significant Effect				
	50*	5.702	2.502	0.037	4	0.0002	CDF	Significant Effect				
<b>ANOVA Table</b>												
Source	Sum Squares		Mean Square		DF	F Stat		P-Value	Decision( $\alpha:5\%$ )			
Between	0.02286096		0.004572191		5	13.73		0.0001	Significant Effect			
Error	0.003997013		0.0003330844		12							
Total	0.02685797				17							
<b>Distributional Tests</b>												
Attribute	Test		Test Stat	Critical	P-Value	Decision( $\alpha:1\%$ )						
Variances	Bartlett Equality of Variance		9.291	15.09	0.0980	Equal Variances						
Variances	Mod Levene Equality of Variance		1.641	8.746	0.2808	Equal Variances						
Variances	Levene Equality of Variance		3.778	5.064	0.0275	Equal Variances						
Distribution	Shapiro-Wilk W Normality		0.9376	0.8546	0.2637	Normal Distribution						
Distribution	Kolmogorov-Smirnov D		0.184	0.2344	0.1086	Normal Distribution						
Distribution	D'Agostino Skewness		1.599	2.576	0.1098	Normal Distribution						
Distribution	Anderson-Darling A2 Normality		0.6187	3.878	0.1082	Normal Distribution						
<b>Frond Weight Summary</b>												
C-mg ai/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect	
0	Negative Control	3	0.1703	0.08341	0.2573	0.1581	0.1431	0.2098	0.0202	20.54%	0.0%	
3.125		3	0.1749	0.1604	0.1895	0.1726	0.1706	0.1816	0.003383	3.35%	-2.7%	
6.25		3	0.185	0.1714	0.1986	0.1825	0.1812	0.1913	0.003172	2.97%	-8.61%	
12.5		3	0.1628	0.1169	0.2086	0.1567	0.1481	0.1835	0.01066	11.34%	4.44%	
25		3	0.118	0.1029	0.1332	0.1159	0.1133	0.1249	0.003514	5.16%	30.7%	
50		3	0.08537	0.04013	0.1306	0.0847	0.0675	0.1039	0.01051	21.33%	49.88%	

## OCSPP 850.4400 Aquatic Vascular Plant

Stillmeadow, Inc.

Analysis ID: 19-4868-2227

Endpoint: Frond Weight

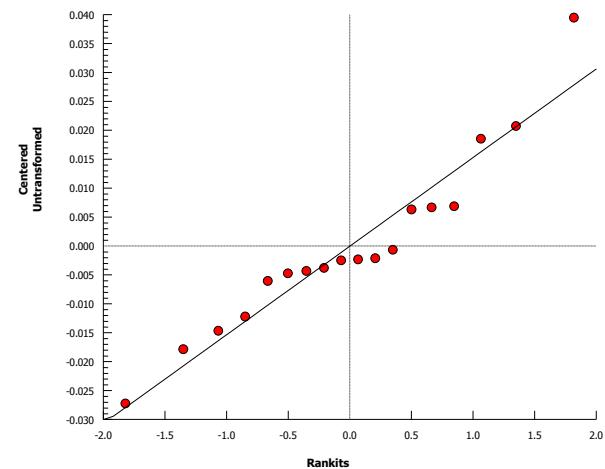
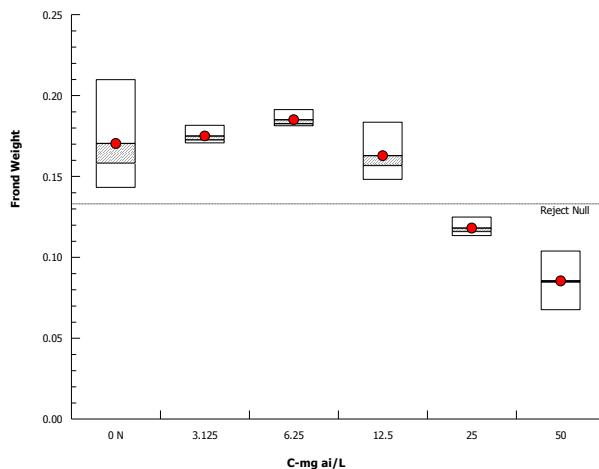
CETIS Version: CETISv1.8.7

Analyzed: 11 Feb-14 11:57

Analysis: Parametric-Control vs Treatments

Official Results: Yes

## Graphics



# CETIS Analytical Report

Report Date: 11 Feb-14 12:00 (p 11 of 12)  
 Test Code: 079701 49044001 | 01-3500-9253

## OCSPP 850.4400 Aquatic Vascular Plant

Stillmeadow, Inc.

Analysis ID:	05-7312-0626	Endpoint:	Frond Weight	CETIS Version:	CETISv1.8.7				
Analyzed:	11 Feb-14 11:57	Analysis:	Parametric-Control vs Ord.Treatments	Official Results:	Yes				
Batch ID:	03-7100-1831	Test Type:	Lemna Growth (7-d)	Analyst:					
Start Date:	05 Nov-12	Protocol:	OCSPP 850.4400 Aquatic Plant (Lemna)	Diluent:	Lemna Medium, 20X-AAP				
Ending Date:	21 Nov-12	Species:	Lemna Gibba	Brine:					
Duration:	16d 0h	Source:	Lab In-House Culture	Age:					
Data Transform	Zeta	Alt Hyp	Trials	Seed	PMSD	NOEL	LOEL	TOEL	TU
Untransformed	NA	C > T	NA	NA	16.9%	12.5	25	17.68	

### Williams Multiple Comparison Test

Control	vs	C-mg ai/L	Test Stat	Critical	MSD	DF	P-Value	P-Type	Decision( $\alpha:5\%$ )
Negative Control		3.125	-0.3087	1.782	0.027	4	>0.05	CDF	Non-Significant Effect
		6.25	-0.6465	1.873	0.028	4	>0.05	CDF	Non-Significant Effect
		12.5	0.5078	1.903	0.028	4	>0.05	CDF	Non-Significant Effect
		25*	3.51	1.918	0.029	4	<0.05	CDF	Significant Effect
		50*	5.702	1.927	0.029	4	<0.05	CDF	Significant Effect

### ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision( $\alpha:5\%$ )
Between	0.02286096	0.004572191	5	13.73	0.0001	Significant Effect
Error	0.003997013	0.0003330844	12			
Total	0.02685797		17			

### Distributional Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision( $\alpha:1\%$ )
Variances	Bartlett Equality of Variance	9.291	15.09	0.0980	Equal Variances
Variances	Mod Levene Equality of Variance	1.641	8.746	0.2808	Equal Variances
Variances	Levene Equality of Variance	3.778	5.064	0.0275	Equal Variances
Distribution	Shapiro-Wilk W Normality	0.9376	0.8546	0.2637	Normal Distribution
Distribution	Kolmogorov-Smirnov D	0.184	0.2344	0.1086	Normal Distribution
Distribution	D'Agostino Skewness	1.599	2.576	0.1098	Normal Distribution
Distribution	Anderson-Darling A2 Normality	0.6187	3.878	0.1082	Normal Distribution

### Frond Weight Summary

C-mg ai/L	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Negative Control	3	0.1703	0.08341	0.2573	0.1581	0.1431	0.2098	0.0202	20.54%	0.0%
3.125		3	0.1749	0.1604	0.1895	0.1726	0.1706	0.1816	0.003383	3.35%	-2.7%
6.25		3	0.185	0.1714	0.1986	0.1825	0.1812	0.1913	0.003172	2.97%	-8.61%
12.5		3	0.1628	0.1169	0.2086	0.1567	0.1481	0.1835	0.01066	11.34%	4.44%
25		3	0.118	0.1029	0.1332	0.1159	0.1133	0.1249	0.003514	5.16%	30.7%
50		3	0.08537	0.04013	0.1306	0.0847	0.0675	0.1039	0.01051	21.33%	49.88%

# CETIS Analytical Report

Report Date: 11 Feb-14 12:00 (p 12 of 12)  
Test Code: 079701 49044001 | 01-3500-9253

## OCSPP 850.4400 Aquatic Vascular Plant

Stillmeadow, Inc.

Analysis ID: 05-7312-0626

Endpoint: Frond Weight

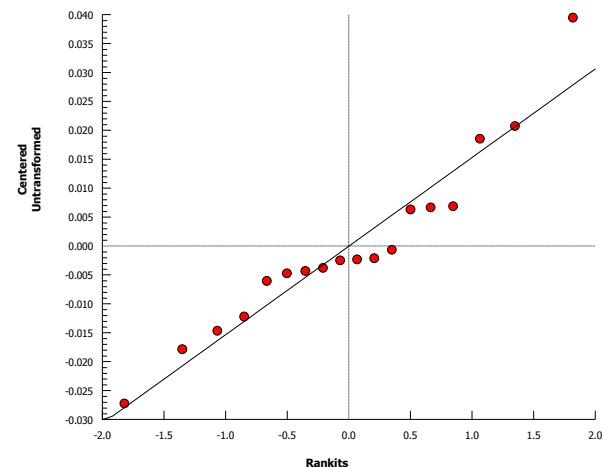
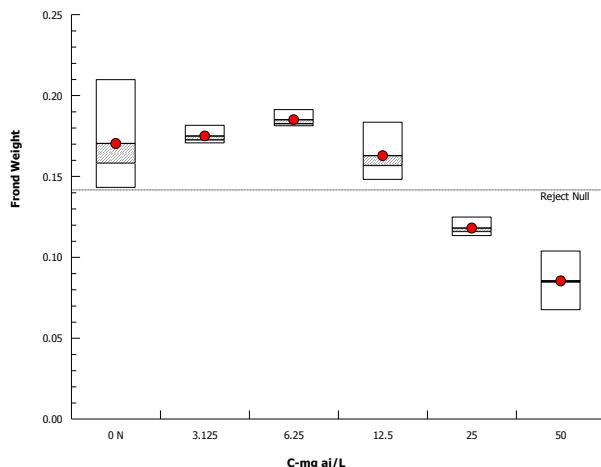
CETIS Version: CETISv1.8.7

Analyzed: 11 Feb-14 11:57

Analysis: Parametric-Control vs Ord.Treatments

Official Results: Yes

### Graphics



# CETIS Analytical Report

Report Date: 11 Feb-14 12:06 (p 1 of 6)  
 Test Code: 079701 49044001 | 01-3500-9253

## OCSPP 850.4400 Aquatic Vascular Plant

Stillmeadow, Inc.

Analysis ID:	10-1825-1279	Endpoint:	Frond Number Growth Rate	CETIS Version:	CETISv1.8.7
Analyzed:	11 Feb-14 11:57	Analysis:	Nonlinear Regression	Official Results:	Yes
Batch ID:	03-7100-1831	Test Type:	Lemna Growth (7-d)	Analyst:	
Start Date:	05 Nov-12	Protocol:	OCSPP 850.4400 Aquatic Plant (Lemna)	Diluent:	Lemna Medium, 20X-AAP
Ending Date:	21 Nov-12	Species:	Lemna Gibba	Brine:	
Duration:	16d 0h	Source:	Lab In-House Culture	Age:	

### Non-Linear Regression Options

Model Function	X Transform	Y Transform	Weighting Function	PTBS Function
3P Cumulative Log-Normal EV [Y=A*(1- Φ(log(X/D)/C))]	None	None	Poisson [W=1/Y]	Off [Y*=Y]

### Regression Summary

Iters	Log LL	AICc	BIC	Adj R2	Optimize	F Stat	Critical	P-Value	Decision(α:5%)
22	-10.72	29.16	30.12	0.7000	Yes	1.31	3.49	0.3165	Non-Significant Lack of Fit

### Point Estimates

Level	mg ai/L	95% LCL	95% UCL
IC5	16.78	N/A	24.83
IC10	22.97	11.48	31.66
IC25	38.81	31.81	46.09
IC50	69.51	50.07	96.52

### Regression Parameters

Parameter	Estimate	Std Error	95% LCL	95% UCL	t Stat	P-Value	Decision(α:5%)
A	0.2739	0.007869	0.2585	0.2894	34.81	<0.0001	Significant Parameter
C	0.8641	0.2819	0.3116	1.417	3.065	0.0079	Significant Parameter
D	69.51	10.97	48	91.02	6.334	<0.0001	Significant Parameter

### ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Model	0.085302	0.085302	1	41.66	<0.0001	Significant
Lack of Fit	0.007577	0.002526	3	1.31	0.3165	Non-Significant
Pure Error	0.023135	0.001928	12			
Residual	0.030712	0.002047	15			

### Residual Analysis

Attribute	Method	Test Stat	Critical	P-Value	Decision(α:5%)
Goodness-of-Fit	Pearson Chi-Sq GOF	0.03071	25	1.0000	Non-Significant Heterogeneity
	Likelihood Ratio GOF	0.03045	25	1.0000	Non-Significant Heterogeneity
Variances	Bartlett Equality of Variance	4.527	11.07	0.4763	Equal Variances
	Mod Levene Equality of Variance	0.3995	4.387	0.8335	Equal Variances
Distribution	Shapiro-Wilk W Normality	0.9471	0.8965	0.3818	Normal Distribution
	Anderson-Darling A2 Normality	0.3668	2.492	0.4372	Normal Distribution

### Frond Number Growth Rate Summary

C-mg ai/L	Control Type	Count	Calculated Variate						
			Mean	Min	Max	Std Err	Std Dev	CV%	%Effect
0	Negative Control	3	0.2913	0.278	0.315	0.01186	0.02055	7.05%	0.0%
3.125		3	0.276	0.266	0.284	0.005292	0.009165	3.32%	5.26%
6.25		3	0.2577	0.248	0.267	0.005487	0.009504	3.69%	11.56%
12.5		3	0.2587	0.245	0.281	0.01126	0.0195	7.54%	11.21%
25		3	0.2477	0.212	0.269	0.01795	0.03109	12.55%	14.99%
50		3	0.1763	0.157	0.207	0.01551	0.02686	15.23%	39.47%

# CETIS Analytical Report

Report Date: 11 Feb-14 12:06 (p 2 of 6)  
 Test Code: 079701 49044001 | 01-3500-9253

## OCSPP 850.4400 Aquatic Vascular Plant

Stillmeadow, Inc.

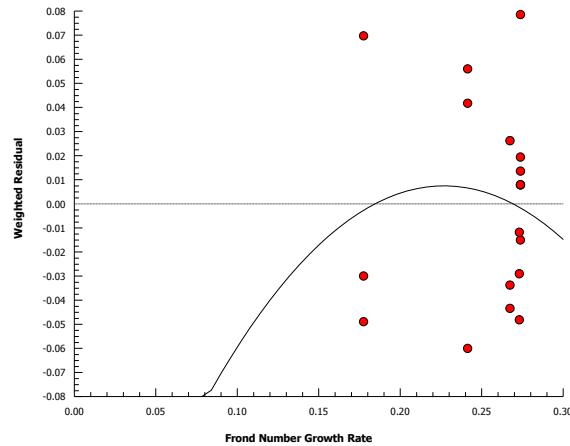
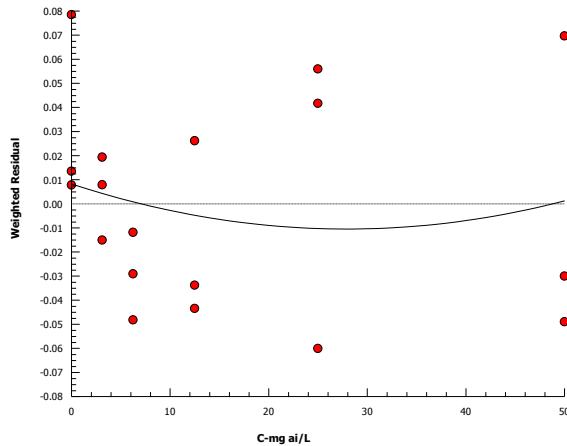
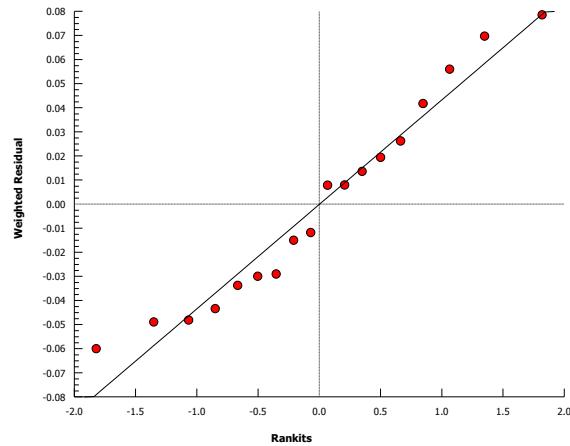
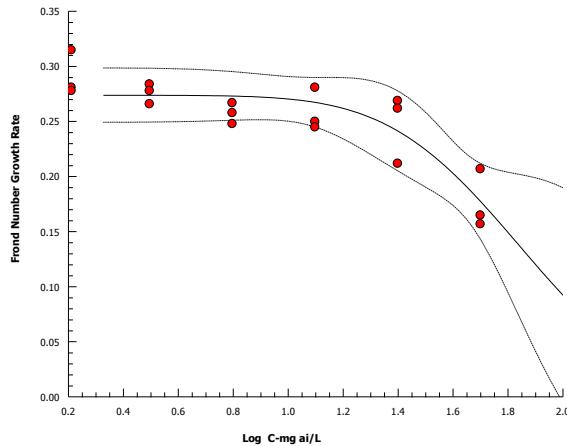
Analysis ID: 10-1825-1279  
 Analyzed: 11 Feb-14 11:57

Endpoint: Frond Number Growth Rate  
 Analysis: Nonlinear Regression

CETIS Version: CETISv1.8.7  
 Official Results: Yes

### Graphics

3P Cumulative Log-Normal EV [Y=A\*(1- Φ(log(X/D)/C))]



# CETIS Analytical Report

Report Date: 11 Feb-14 12:06 (p 3 of 6)  
 Test Code: 079701 49044001 | 01-3500-9253

## OCSPP 850.4400 Aquatic Vascular Plant

Stillmeadow, Inc.

Analysis ID:	04-1108-0388	Endpoint:	Frond Number	CETIS Version:	CETISv1.8.7
Analyzed:	11 Feb-14 11:57	Analysis:	Nonlinear Regression	Official Results:	Yes
Batch ID:	03-7100-1831	Test Type:	Lemna Growth (7-d)	Analyst:	
Start Date:	05 Nov-12	Protocol:	OCSPP 850.4400 Aquatic Plant (Lemna)	Diluent:	Lemna Medium, 20X-AAP
Ending Date:	21 Nov-12	Species:	Lemna Gibba	Brine:	
Duration:	16d 0h	Source:	Lab In-House Culture	Age:	

### Non-Linear Regression Options

Model Function	X Transform	Y Transform	Weighting Function	PTBS Function
3P Cumulative Log-Normal EV [Y=A*(1- Φ(log(X/D)/C))]	None	None	Poisson [W=1/Y]	Off [Y*=Y]

### Regression Summary

Iters	Log LL	AICc	BIC	Adj R2	Optimize	F Stat	Critical	P-Value	Decision(α:5%)
12	-72.9	153.5	154.5	0.6559	Yes	2.216	3.49	0.1389	Non-Significant Lack of Fit

### Point Estimates

Level	mg ai/L	95% LCL	95% UCL
IC5	1.213	N/A	4.678
IC10	2.457	N/A	6.174
IC25	7.994	3.982	13.97
IC50	29.65	19.84	44.32

### Regression Parameters

Parameter	Estimate	Std Error	95% LCL	95% UCL	t Stat	P-Value	Decision(α:5%)
A	89.55	8.6	72.7	106.4	10.41	<0.0001	Significant Parameter
C	1.943	0.5671	0.832	3.055	3.427	0.0037	Significant Parameter
D	29.65	9.553	10.93	48.38	3.104	0.0073	Significant Parameter

### ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Model	91.16125	91.16125	1	34.41	<0.0001	Significant
Lack of Fit	14.16826	4.722754	3	2.216	0.1389	Non-Significant
Pure Error	25.57101	2.130917	12			
Residual	39.73927	2.649285	15			

### Residual Analysis

Attribute	Method	Test Stat	Critical	P-Value	Decision(α:5%)
Goodness-of-Fit	Pearson Chi-Sq GOF	39.74	25	0.0005	Significant Heterogeneity
	Likelihood Ratio GOF	39.12	25	0.0006	Significant Heterogeneity
Variances	Bartlett Equality of Variance	2.945	11.07	0.7084	Equal Variances
	Mod Levene Equality of Variance	0.5235	4.387	0.7528	Equal Variances
Distribution	Shapiro-Wilk W Normality	0.9161	0.8965	0.1101	Normal Distribution
	Anderson-Darling A2 Normality	0.5718	2.492	0.1416	Normal Distribution

### Frond Number Summary

C-mg ai/L	Control Type	Count	Calculated Variate						
			Mean	Min	Max	Std Err	Std Dev	CV%	%Effect
0	Negative Control	3	92.33	74	113	11.32	19.6	21.23%	0.0%
3.125		3	77	65	84	6.028	10.44	13.56%	16.61%
6.25		3	61	56	66	2.887	5	8.2%	33.94%
12.5		3	65	57	74	4.933	8.544	13.14%	29.6%
25		3	57	41	67	8.083	14	24.56%	38.27%
50		3	29.67	24	39	4.702	8.145	27.45%	67.87%

# CETIS Analytical Report

Report Date: 11 Feb-14 12:06 (p 4 of 6)  
 Test Code: 079701 49044001 | 01-3500-9253

## OCSPP 850.4400 Aquatic Vascular Plant

Stillmeadow, Inc.

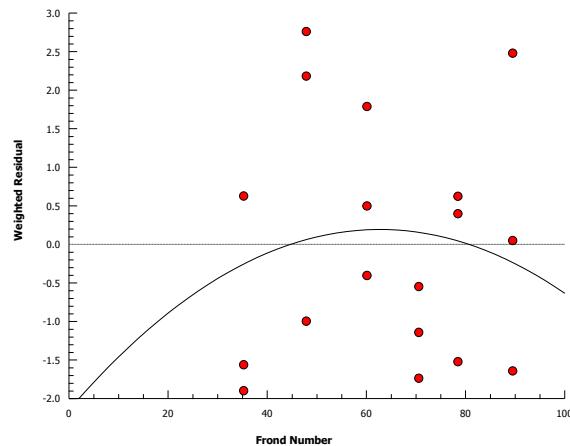
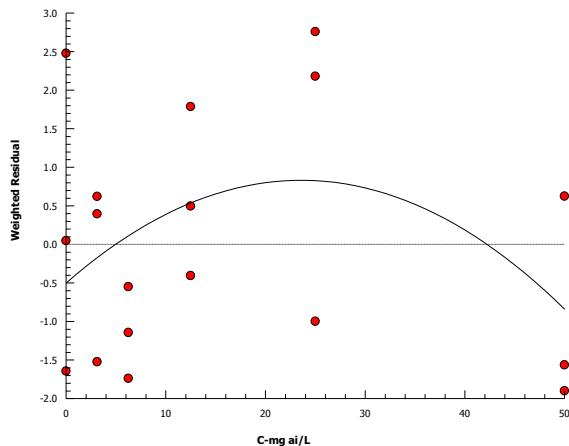
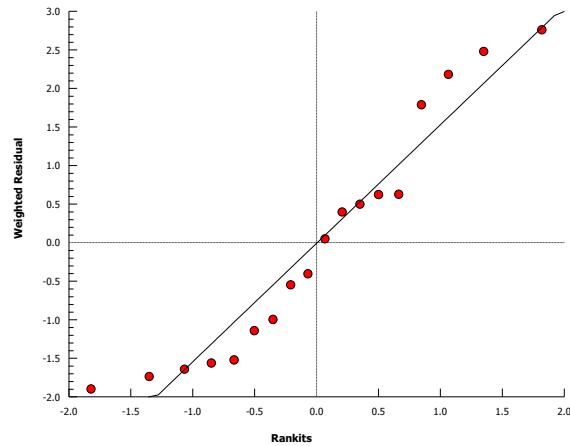
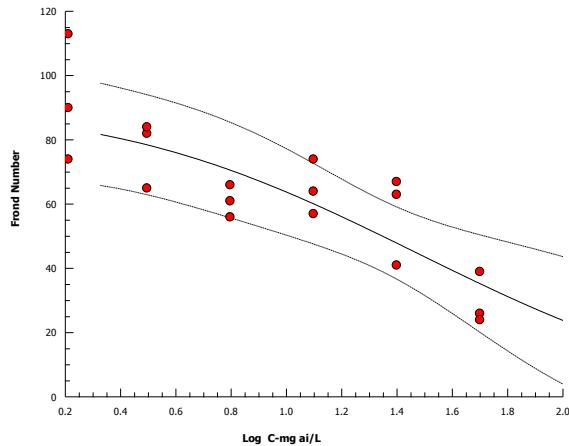
Analysis ID: 04-1108-0388  
 Analyzed: 11 Feb-14 11:57

Endpoint: Frond Number  
 Analysis: Nonlinear Regression

CETIS Version: CETISv1.8.7  
 Official Results: Yes

### Graphics

3P Cumulative Log-Normal EV [Y=A\*(1- Φ(log(X/D)/C))]



# CETIS Analytical Report

Report Date: 11 Feb-14 12:06 (p 5 of 6)  
 Test Code: 079701 49044001 | 01-3500-9253

## OCSPP 850.4400 Aquatic Vascular Plant

Stillmeadow, Inc.

Analysis ID:	10-8444-5970	Endpoint:	Frond Weight	CETIS Version:	CETISv1.8.7
Analyzed:	11 Feb-14 11:57	Analysis:	Nonlinear Regression	Official Results:	Yes
Batch ID:	03-7100-1831	Test Type:	Lemna Growth (7-d)	Analyst:	
Start Date:	05 Nov-12	Protocol:	OCSPP 850.4400 Aquatic Plant (Lemna)	Diluent:	Lemna Medium, 20X-AAP
Ending Date:	21 Nov-12	Species:	Lemna Gibba	Brine:	
Duration:	16d 0h	Source:	Lab In-House Culture	Age:	

### Non-Linear Regression Options

Model Function	X Transform	Y Transform	Weighting Function	PTBS Function
3P Cumulative Log-Normal EV [Y=A*(1- Φ(log(X/D)/C))]	None	None	Poisson [W=1/Y]	Off [Y*=Y]

### Regression Summary

Iters	Log LL	AICc	BIC	Adj R2	Optimize	F Stat	Critical	P-Value	Decision(α:5%)
16	-7.722	23.16	24.11	0.7873	Yes	0.8743	3.49	0.4815	Non-Significant Lack of Fit

### Point Estimates

Level	mg ai/L	95% LCL	95% UCL
IC5	7.941	N/A	12.72
IC10	11.66	5.347	17.05
IC25	22.15	16.32	28.58
IC50	45.2	36.91	55.35

### Regression Parameters

Parameter	Estimate	Std Error	95% LCL	95% UCL	t Stat	P-Value	Decision(α:5%)
A	0.1784	0.007967	0.1627	0.194	22.39	<0.0001	Significant Parameter
C	1.057	0.255	0.5575	1.557	4.146	0.0009	Significant Parameter
D	45.2	5.27	34.87	55.53	8.576	<0.0001	Significant Parameter

### ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Model	0.146034	0.146034	1	64.94	<0.0001	Significant
Lack of Fit	0.006050	0.002017	3	0.8743	0.4815	Non-Significant
Pure Error	0.027681	0.002307	12			
Residual	0.033731	0.002249	15			

### Residual Analysis

Attribute	Method	Test Stat	Critical	P-Value	Decision(α:5%)
Goodness-of-Fit	Pearson Chi-Sq GOF	0.03373	25	1.0000	Non-Significant Heterogeneity
	Likelihood Ratio GOF	0.03361	25	1.0000	Non-Significant Heterogeneity
Variances	Bartlett Equality of Variance	9.19	11.07	0.1017	Equal Variances
	Mod Levene Equality of Variance	1.867	4.387	0.2345	Equal Variances
Distribution	Shapiro-Wilk W Normality	0.9702	0.8965	0.8022	Normal Distribution
	Anderson-Darling A2 Normality	0.2266	2.492	0.8483	Normal Distribution

### Frond Weight Summary

C-mg ai/L	Control Type	Count	Calculated Variate						
			Mean	Min	Max	Std Err	Std Dev	CV%	%Effect
0	Negative Control	3	0.1703	0.1431	0.2098	0.0202	0.03499	20.54%	0.0%
3.125		3	0.1749	0.1706	0.1816	0.003383	0.005859	3.35%	-2.7%
6.25		3	0.185	0.1812	0.1913	0.003172	0.005494	2.97%	-8.61%
12.5		3	0.1628	0.1481	0.1835	0.01066	0.01846	11.34%	4.44%
25		3	0.118	0.1133	0.1249	0.003514	0.006087	5.16%	30.7%
50		3	0.08537	0.0675	0.1039	0.01051	0.01821	21.33%	49.88%

## OCSPP 850.4400 Aquatic Vascular Plant

Stillmeadow, Inc.

Analysis ID: 10-8444-5970  
 Analyzed: 11 Feb-14 11:57

Endpoint: Frond Weight  
 Analysis: Nonlinear Regression

CETIS Version: CETISv1.8.7  
 Official Results: Yes

## Graphics

3P Cumulative Log-Normal EV [Y=A\*(1- Φ(log(X/D)/C))]

